Cancer Target Discovery and Development (CTD^2)

Specific Aims

Institution: The Translational Genomics Research Institute (TGEN)

Recent profiling efforts of human cancers such as TCGA, ICGC, TARGET, and others portray detailed and varied aberrations in the molecular genetics of cancers. Such thorough characterization has offered a biological basis for dividing histologically homogeneous tumors into molecular subtypes with clinically- meaningful insight. For glioblastoma, informatics outcomes based on these profiles have contributed to subtypes of predictable and distinct survival outcomes, as well as patterns or rates of progression, and dissemination behaviors. These prognostic advances have ushered in an expectation that underlying molecular drivers of malignant phenotypes within histologically-grouped tumors can be identified, and that such molecular profiles will offer an opportunity to both discover novel therapeutic targets as well as to deploy molecular biomarkers which would prospectively align specific subgroups with optimally effective (or decidedly ineffective) targeted therapeutics. As large-scale profiling efforts expand to more than 20 additional tumor types, systematic processes are highly desirable by which novel, actionable targets are identified, validated, and developed into discovery tools for new therapies.

As a node in the Cancer Target Discovery & Development (CTD^2) Network, our team includes TGen's In Silico Research Center of Excellence (ISRCE) (SAIC-Frederick contract) has significant knowledge of The Cancer Genome Atlas on Glioblastoma Multiforme (GBM), and brings a wealth of bioinformatic tools for database mining and in silico target identification. Expertise includes development and use of 54 molecularly-profiled human orthotopic primary GBM xenografts, and analysis of their *in vivo* phenotypes. Systems biology expertise is provided by the Van Andel Research Institute (VARI). The informatics capabilities and xenograft models serve as resources for subsequent functional target and pathway identification and validation at Sanford-Burnham Medical Research Institute (SBMRI). SBMRI's Center for Chemical Genomics [Comprehensive Center in NIH's Molecular Libraries Probe Production Centers Network and NCI's Chemical Biology Consortium]] enables robust RNAi and small-molecule-based high-throughput assay development and screening for large-scale functional validation of targets and pathways. Thomson Reuters will provide an expertise in systems biology of cancer, bioinformatics and pathway analysis of OMICs data and causal network analysis.

The **Specific Aims** of the Project are:

Aim 1. Deploy bioinformatics tools for novel target discovery in glioblastoma. i) Mine large profiling datasets of tumors using commercial and custom algorithms for *in silico* target identification in subgroups of tumors; ii) sub-classify tumor types into distinct groups each with its distinct candidate novel therapeutic target. In both instances apply

curated, distinct, and clinically-meaningful behaviors matched with preclinical *in vivo* and *in vitro* models.

Aim 2. Functionally validate novel targets in the various GBM subgroups by using **RNAi-based high- throughput approaches**. Drawing from expansive preclinical models, develop cell-based HT assays for evaluation of "Hallmarks of Cancer" *in vitro* in the molecularly-profiled subgroups of GBM. Use these assays in focused shRNA testing to functionally validate *in silico*-identified targets in the relevant GBM subgroups. Perform broader pathway-based RNAi screening to enable systems biology assessment of the GBM subgroups. Such assessment will be used to validate and optimize the *in silico* tumor sub-groupings in an iterative manner, as well as to functionally identify unique molecular vulnerabilities in an unbiased manner. Utilize engineered, inducible shRNA knockdown xenografts for *in vivo* biological validation of "hallmark" endpoints.

Aim 3. Functionally validate novel targets in the various GBM subgroups **using bioactive small molecule high-throughput approaches**. Validate informatically-identified targets using chemical approaches. This includes using small molecules to validate specific targets and pathways, screening with a cancer signaling pathway inhibitor library, and testing of compound function *in vivo*. Additionally, if specific compounds against most promising shRNA-identified targets are not available, HTS campaigns will be undertaken to identify suitable chemical probes. This approach will enable rapid bridging of the gap from *in vitro* target validation to the identification of chemical probes and prototype therapeutics.

The project **significantly** pursues targets that drive the lethal hallmarks of glioblastoma, is **innovative** in its early inclusion of preclinical models to render targets "actionable" and deploys an **approach** that is systematic (workflow-based), validating, iterative, and uses state-of-the-art competencies.